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Evaluation of a previous mass campaign against endemic syphilis in Bosnia and Herzegovina

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It is now almost 30 years since penicillin was introduced to combat the endemic treponematoses of childhood, prevalent in rural areas of many developing countries. Preparations such as PAM and benzathine penicillin were shown to produce effective blood and tissue concentrations of 4 days' to 3 weeks' duration after single intramuscular injections. Long-acting treatment thus became available which could be used on clinical as well as epidemiological indications, and organized community-wide campaigns could be undertaken in endemic treponematoses areas. This development gave hope that disease could be eliminated by the wide use of a drug alone.

On the basis of pilot studies of yaws in Haiti (Levitan, Rodriguez, Jacobs, Petrus, and Durand, 1953), endemic childhood syphilis in Yugoslavia (Grin, 1953), and pinta in Mexico (Edmundson, Lopez, Rico, and Olansky, 1953), mass penicillin campaigns were undertaken by health administrations in 46 countries in the context of the WHO Treponematoses Programme. In the first decade national and international attention was focused on the setting up of a programme and on control of disease. With declining clinical prevalence, orientation turned towards concurrent epidemiological surveillance. In the last few years sero-epidemiological studies of the changing pattern of disease and infection became possible and a methodology for long-term surveillance of endemic treponematoses was developed in the frame of the activities of the World Health Organization. The present paper is concerned with the application of this methodology and the study of 'disappearing' disease in regard to the endemic treponematosis of childhood, known as 'endemic non-venereal syphilis', previously prevalent in the Federal Republic of Bosnia-Herzegovina, Yugoslavia.

It may first be useful to recall that endemic syphilis is a disease acquired by non-venereal contact, predominantly in childhood, characterized by infectious mucous membrane and relapsing skin lesions in the first years of its natural course. In adolescence and adult life, late, sometimes incapacitating and mutilating, lesions of skin, bones, and joints may develop. Cardiovascular and nervous system involvement are rare, and when present are mostly mild and non-progressive.

Early latency and late latency are much more frequent than clinical disease in endemic areas. These latent cases, as well as those with clinical lesions, are seroreactive in tests using lipoidal and treponemal antigens (e.g. the Wassermann reaction (WR), the Venereal Disease Research Laboratory (VDRL) test, the fluorescent treponemal antibody (FTA) test, and the *Treponema pallidum* immobilization (TPI) test. Serological response to therapy is related to the duration of the infection. Seroreactivity may remain in endemic syphilis as in other treponematoses throughout life in untreated persons and often also in persons adequately treated in the later stages of the disease.

Secondly, it may be recalled that, when therapy of the treponematoses depended on multiple injections of arsenicals and/or bismuth, mass campaigns were nevertheless attempted against the endemic treponematoses in several countries. Treatment surveys and re-surveys were undertaken in rural populations and information was obtained on the nature, extent, distribution, and pattern of disease. Such epidemiological data were, for example, collected on yaws in

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Western Pacific islands as early as 1923 and in subsequent years (Buxton, 1928; Lambert, 1936), and in Africa (Harding, 1949), as well as on endemic syphilis in Yugoslavia (Grin, 1937; Kogoj and Vuletic, 1939). However, the epidemiological concept that treatment was necessary for symptomless household contacts and presumed latent cases in addition to clinical cases had as yet not evolved. Demographic aspects were not taken adequately into account in early campaigns, nor was the epidemiological importance of obtaining complete coverage of the total population recognized. Such quantitative aspects are today held to be essential elements of mass campaigns and of the methodology of epidemiological surveillance.

In general, several periodical re-surveys (RS) are undertaken after the initial treatment survey (ITS) of mass campaigns against the endemic treponematoses. From the findings in the mass campaigns and the concurrent surveillance studies of endemic treponematoses certain questions arise:

> *Can early infectious clinical disease actually be eliminated, and can transmission of infection be interrupted through the wide application of a drug, utilized in accordance with the epidemiological characteristics of the disease?*

The endemic syphilis of Bosnia and the mass-treatment campaign, 1948-1955

Since the first published description of endemic syphilis ('non-venereal endemic syphilis') in Bosnia-Herzegovina by Glück (1888), the origin, extent, and changing pattern of this disease have been the subject of many national and international studies. These are not reviewed in the present report, which refers only to recent investigations of this endemic disease after a penicillin mass treatment programme which was begun in 1948. The programme has been described in some detail by Grin (1953, 1960), Clark (1950), Guthe and Luger (1966), and others. Grin also described the changing socioeconomic conditions of the population in the endemic areas as well as the relevant cultural and other background features which affect the epidemiology and control of endemic treponematosis.

The campaign, which was conducted between 1948 and 1955 by the health administration of the Republic of Bosnia-Herzegovina and the federal health authorities of Yugoslavia, with technical and other assistance from the World Health Organization (WHO) and the United Nations Children's Fund (UNICEF), aimed at the eradication of this endemic treponematosis of childhood which had been prevalent in large areas of the Republic since the 18th century (Fig. 1).

FIG. 1 *Prevalence of endemic syphilis in Bosnia and Herzegovina*, 1951

The methodology used in the campaign has been described in the publications mentioned as well as in reports by WHO Expert Committees (WHO, 1953, 1960). Briefly, the methodology was based on an initial examination of the population concerned at ITS, with treatment of overt clinical and latent cases and their household contacts, followed by periodical RS of the populations at risk, in both instances using defined dosage schedules of long-acting procaine penicillin G in oil with aluminium monostearate (PAM).

The coverage of the population in the ITS of the mass campaign—during which more than 600,000 people were examined—ranged from 50·1 to 99·6 per cent. of the enumerated population in different areas (Table I and Fig. 2). An average of 79·2 per cent. of the population at risk was examined. The clinical and serological findings at the ITS and succesive RSs in the population covered by the mass campaign 1948–1955 and on subsequent surveillance (1956–1967) are presented in Table II(a, b, c, d, e) (overleaf). At ITS, infectious lesions ranged from 0 to 4·4 per cent. and seroreactivity from 0·75 to 16·6 per cent. in the different administrative units* as a whole. There were great variations in the endemicity of the disease between the different smaller units and villages in one Srez and between the different Srez. In some villages the incidence of infectious lesions was as high as 14 per cent. (as in Sokol) and that of seroreactors was more than 50 per cent. (as in Brushnica). This pattern is characteristic of endemic syphilis, and foci of high prevalence are often found next to areas of very low prevalence. This aspect is illustrated in Fig. 3.

*The local name for an administrative unit is Srez.

TABLE I *Population coverage at the initial survey (ITS), 1948–56*

Srez				*Population in villages covered by ITS*			
No. (1)	*Name* (2)	*Estimated population 1953* (3)	*Number of MNOs[a] covered by ITS* (4)	*No. enumerated* (5)	*Per cent. of estimated population* (6)	*No. examined* (7)	*Per cent. of enumerated population* (8)
01	Bijeljina	86,800	6	22,979	26·5	18,865	82·1
02	Brčko	65,300	8	19,035	29·2	16,713	87·8
03	Bosanska	40,600	6	40,161	98·9	38,722	96·4
04	Bihač	39,200	7	38,124	97·3	33,755	88·5
05	Bugojino	59,100	12	40,534	68·6	34,028	83·9
06	Cazin	36,300	13	33,472	92·2	26,114	78·0
07	Doboj	56,500	1	3,419	6.1	2,698	78·9
08	Foča	39,400	6	31,934	81·1	25,884	81·1
09	Gračanica	47,300	12	45,888	97·0	32,071	69·9
10	Gardačac	43,200	13	42,680	98·8	32,104	75·2
11	Kladanj	18,700	9	14,587	78·0	11,179	76·6
12	Lopare	36,000	10	31,660	87·9	24,601	77·7
13	Maglaj	28,200	1	9,180	32·6	7,301	79·5
14	Modrica	39,200	1	1,458	3·7	1,240	85·0
15	Prozor	18,400	1	6,405	34·8	4,505	70·3
16	Prijedor	74,400	1	7,812	10·5	6,400	81·9
17	Sokolac	16,200	1	295	1·8	205	69·5
18	Srebrenica	46,700	13	33,036	70·7	26,987	81·7
19	Teslic	39,900	3	4,463	11·2	3,725	83·5
20	Tešanj	26,900	9	24,667	91·7	19,661	79·7
21	Tuzla	121,600	23	99,381	81·7	81,388	81·9
22	Travnik	72,600	6	60,573	83·4	49,274	81·3
23	Zavidovići	44,200	3	12,332	27·9	10,623	86·2
24	Zenica	54,400	16	50,442	92·7	27,923	55·4
25	Zvornik	53,700	15	48,468	90·3	39,551	81·6
26	Vares	22,400	1	764	3·4	694	90·8
27	Visoko	65,100	14	48,662	74·7	34,855	71·6
28	Vlasenica	30,900	10	28,459	92·1	18,779	66·0
29	Velika Kladuša	23,400	4	23,322	99·7	23,240	99·6
30	Posušje	22,400	1	3,653	16·3	1,829	50·1
Total		1,369,000	226	827,825	60·5	654,914	79·1

[a]MNO = Administrative unit

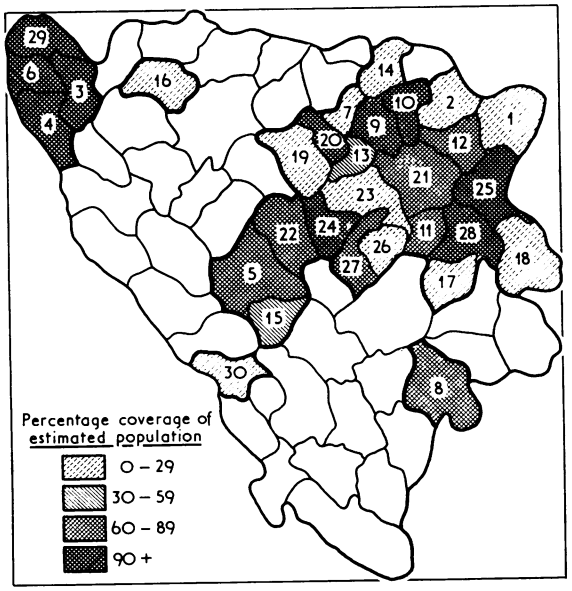

FIG. 2 *Proportion of population covered by the initial treatment survey (ITS) in each Srez. The numbers correspond to the Srez code as given in Table I*

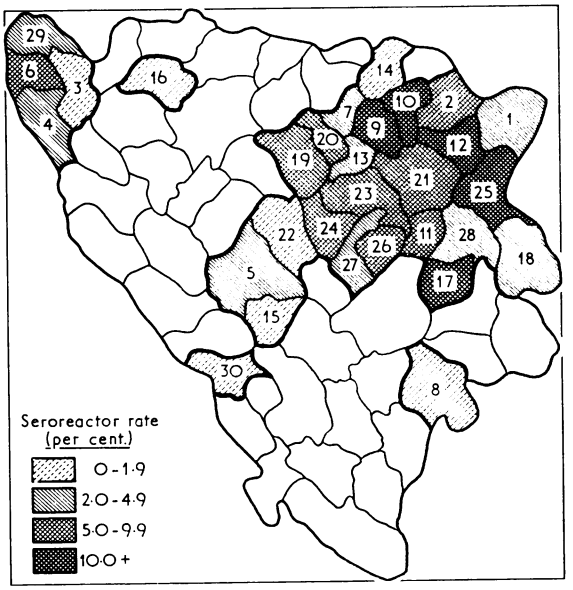

FIG. 3 *Average seroreactor rate in the initial treatment survey (ITS) in each Srez*

TABLE II *Clinical and serological findings in population covered by mass campaign*

Survey		*Initial treatment survey*					*First re-survey* (1950-54)				
Srez			*Infectious cases*		*Seroreactivity*			*Infectious cases*		*Seroreactivity*	
Code No.	*Name*	*Population examined*	*No.*	*Infectious Rate per cent.*	*No. positive*[a]	*Reactor Rate per cent.*	*Population examined*	*No.*	*Infectious Rate per cent.*	*No. positive*[a]	*Re Ra pe*
01	Bijeljina	18,865	29	0·15	448	2·4	1,630	4	0·2	83	5
02	Brčko	16,713	37	0·22	1,458	8·7	6,914	2	0·03	445	6
03	Bosanska	33,722	31	0·08	715	1·8	11,743	0	0	165	1
04	Bihač	33,755	18	0·05	878	2·6	501	0	0	10	2
05	Bugojino	34,028	95	0·28	1,110	3·3	11,059	6	0·05	632	5
06	Cazin	26,114	120	0·46	2,056	7·9	30,052	21	0·07	510	1
07	Doboj	2,698	—	0·00	108	4·0	0	0	0	0	0
08	Foča	25,884	11	0·04	193	0·75	886	0	0	6	0
09	Gračanica	32,071	950	3·0	4,477	14·0	27,558	271	1·0	4,456	16
10	Gradačac	32,104	297	0·93	4,823	15·0	28,595	67	0·2	1,629	5
11	Kladanj	11,179	40	0·36	555	5·0	6,490	4	0·06	143	2
12	Lopare	24,601	221	0·90	2,800	11·4	18,933	31	0·2	2,086	11
13	Maglaj	7,301	14	0·19	215	2·9	1,943	2	0·1	46	2
14	Modrica	1,240	—	0·00	12	0·97	0	0	0	0	0
15	Prozor	4,505	1	0·02	44	0·98	235	0	0	1	0
16	Prijedor	6,400	—	0·00	106	1·7	4,624	0	0	16	0
17	Sokolac	205	9	4·4	34	16·6	0	0	0	0	0
18	Srebrenica	26,987	66	0·24	862	3·2	3,140	7	0·2	146	4
19	Teslic	3,725	13	0·35	230	6·2	1,751	1	0·06	28	1
20	Tešanj	19,661	81	0·41	1,397	7·1	14,075	23	0·2	644	4
21	Tuzla	81,388	469	0·58	5,639	6·9	39,603	53	0·1	1,635	4
22	Travnik	49,274	18	0·04	674	1·4	1,045	0	0	16	1
23	Zavidovići	10,623	31	0·29	597	5·6	2,468	1	0·04	62	2
24	Zenica	27,923	—	0·00	1,701	6·1	829	0	0	6	0
25	Zvornik	39,551	257	0·65	5,219	13·2	38,449	76	0·2	1,184	3
26	Vares	694	4	0·58	60	8·6	0	0	0	0	0
27	Visoko	34,855	17	0·05	1,178	3·4	958	0	0	16	1
28	Vlasenica	18,779	40	0·21	682	3·6	1,541	14	0·9	291	18
29	Velika Kladuša	23,240	27	0·12	630	2·7	581	0	0	5	0
30	Posušje	1,829	—	0·00	20	1·1	154	0	0	0	0
Total		654,914	2,896	0·44	38,921	5·94	255,757	583	0·2	14,261	5·

[a]Including weakly positive

Table II(e) shows that it was necessary to follow up only six Srez with a fourth RS, during which a total of approximately 21,000 people were examined. After 1955, surveillance activities were undertaken in periodical RS in the previously infected areas, either as separate actions or integrated in the activities of a subsequent antimycosis campaign (Grin, 1961). A more detailed picture of the regression of endemic syphilis after the mass penicillin campaigns, not only in the Srez of Bosnia-Herzegovina but also in the villages themselves, is presented in Table III, showing the findings at the ITS and at the last re-survey (LRS) after completion of the campaign in twenty villages situated in nine regions. The complete disappearance of active lesions and the decreased rate of seroreactivity emphasize the effectiveness of the mass campaign. There can be little doubt that the effect has been conspicuous throughout the endemic syphilis areas in Bosnia both as regards clinical cases and seroreactivity, indicating complete eradication of the infection. The WHO Expert Committee on Venereal Infections and Treponematoses (*WHO Technical Report Series* No. 190, 1960) defined two stages of eradication: 'epidemiological eradication' and 'complete eradication'. The former was recognized as the stage at which no indigenous *infectious* case of treponematosis had appeared in the population for 3 consecutive years, in accordance with certain criteria established for casefinding. 'Complete eradication' would require that no *active* case appeared in the population for 3 years and that no seroreactors were found among children under 5 years of age—again in accordance with certain criteria for casefinding. Serological investigations of endemic treponematoses after mass campaigns are thus necessary in order to evaluate the effect of the campaign.

Post mass-campaign evaluation study

An agreement to carry out a representative seroepidemiological study of the endemic treponematoses of Bosnia-Herzegovina 20 years after the beginning of the campaign was signed between the Federal Government of Yugoslavia, the Government of the Republic of Bosnia-Herzegovina, and WHO in 1967. With collaboration between the Institute of Dermato-

d re-survey (1951-54)					*Third re-survey* (1953-54)					*Fourth re-survey* (1953-55)				
	Infectious cases		*Seroreactivity*			*Infectious cases*		*Seroreactivity*			*Infectious cases*		*Seroreactivity*	
ation ned	*No.*	*Infectious Rate per cent.*	*No. positive*[a]	*Reactor Rate per cent.*	*Population examined*	*No.*	*Infectious Rate per cent.*	*No. positive*[a]	*Reactor Rate per cent*	*Population examined*	*No.*	*Infectious Rate per cent.*	*No. positive*[a]	*Reactor Rate per cent.*
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
81	9	0·2	129	3·5	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
66	0	0	3	1·1	0	0	0	0	0	0	0	0	0	0
41	0	0	1	0·2	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
40	58	0·2	1,627	4·8	40,864	30	0·07	841	2·1	11,428	13	0·1	392	3·4
72	12	0·04	623	1·9	692	0	0	17	2·5	0	0	0	0	0
50	3	0·7	23	5·1	319	0	0	3	0·9	360	0	0	8	2·2
52	4	0·02	386	1·6	18,465	2	0·01	304	1·6	2,686	1	0·04	77	2·9
94	0	0	38	5·6	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
11	0	0	66	2·6	1,309	0	0	59	4·5	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
34	0	0	120	4·2	0	0	0	0	0	0	0	0	0	0
15	34	0·1	890	3·4	7,428	1	0·01	223	3·0	2,338	0	0	20	0·9
75	0	0	0	0	0	0	0	0	0	0	0	0	0	0
73	0	0	6	1·3	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
76	24	0·1	530	3·2	6,210	0	0	126	2·0	3,729	2	0·05	91	2·4
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
92	9	9·1	53	5·3	504	1	0·2	43	8·5	916	1	0·1	51	5·6
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
982	153	0·1	4,495	3·0	75,791	34	0·04	1,616	2·1	21,457	17	0·08	639	3·0

TABLE III *Results of the initial survey and last re-survey after completion of the campaign in selected villages**

		Initial survey					*Last re-survey after completion of the campaign*					
			No. of people examined	*Active lesions*		*Seroreactive*		*No. of people examined*	*Active lesions*		*Seroreactive*	
Region	*Village*	*Year*		*Early*	*Late*	*Total*	*Per cent.*	*Year*		*Early*	*Late*	*Total*	*Per cent.*
GRAČANICA:	Doborovci	1948	1,930	128	30	468	24·2	1965	1,512	0	0	24	1·6
	Malesic	1948	1,912	143	30	437	22·9	1961	1,915	0	0	71	3·7
	Sladna	1948	2,586	119	27	617	23·9	1961	2,826	0	0	84	3·0
	Sokol	1948	497	71	12	227	45·7	1960	504	0	0	21	4·2
LOPARE:	Brusnica	1949	328	21	1	192	58.5	1960	353	0	0	1	0·3
	Sibosnica	1949	2,668	44	22	428	16·0	1964	2,432	0	0	63	2·6
ZVORNIK:	Dulici	1949	158	0	0	22	13·9	1958	171	0	0	5	2·9
	Kamenica	1949	589	3	7	85	14·4	1965	469	0	0	7	1·5
	Sapna	1949	3,687	136	53	905	24·5	1958	3,386	0	0	44	1·3
KLADANJ:	Nocajevici	1950	254	6	0	45	17·7	1963	360	0	0	8	2·2
	Tarevo	1950	379	0	0	24	6·3	1964	516	0	0	7	1·4
VLASENICA:	Besici a. Nurici	1950	182	1	1	16	8·8	1967	218	0	0	3	1·4
	Bisina a. Sebiocina	1950	208	2	0	7	3·4	1966	324	0	0	12	3·7
TEŠANJ:	Kosova a. Mosevac	1951	1,095	8	24	211	19·3	1962	1,509	0	0	84	5·6
	Mitrovici	1951	369	0	0	9	2·4	1962	338	0	0	2	0·6
SREBRENICA:	Kravica	1951	763	14	13	108	14·2	1960	383	0	0	18	4·7
	Suceska	1951	1,189	21	14	124	10·4	1957	926	0	0	41	4·4
FOČA:	Kozja Luka	1952	562	2	3	18	3·2	1962	420	0	0	4	1·0
PRIJEDOR:	Kozarac	1953	6,400	0	0	106	1·7	1963	4,624	0	0	16	0·3
Total			25,756	719	237	4,049	15·7		23,186	0	0	515	2·2

*In the seroepidemiological survey 1968-1970 some of the villages shown in this Table fell into the randomly selected sample

Venereology, Sarajevo, WHO headquarters, Geneva, and the WHO Regional Office for Europe, Copenhagen, this study was carried out from 1968 to 1970. It was part of the agreement that the serum collections to be obtained would be utilized also for multiple investigations of infections and conditions other than treponematoses.

OBJECTIVES

These are outlined in the Survey Design and Working Protocol (INT/VDT/70.315).

(*a*) To evaluate the results of the previous mass campaign and the epidemiological trends of endemic treponematoses under the influence of mass treatment and of the rapid improvement of socio-economic conditions;
(*b*) To assess the effect of treatment in the community as a whole;
(*c*) To appraise, retrospectively, the effect of treatment in groups actually subjected to mass campaign measures;
(*d*) To undertake special clinical, laboratory and other examinations of persons still found to be seroreactive by the TPI test in the present survey;
(*e*) To exploit the serum collection obtained for conditions other than endemic treponematosis.*

METHODOLOGY

A representative sample of the population of the previous endemic areas was examined serologically by multiple treponemal and lipoidal antigen tests according to the detailed Survey Design and Working Protocol. The laboratory at the Institute in Sarajevo examined all sera. A subsample of twelve villages provided aliquots of sera which were sent to the WHO Reference Laboratory in Copenhagen for proficiency testing as part of an agreement with the Sarajevo Laboratory. A further proficiency testing study of the TPI test was undertaken between the Sarajevo Laboratory, the VDRL Reference Laboratory, Atlanta, and the State Serum Institute Reference Laboratory, Copenhagen, according to a special protocol established for the purpose.

Sample design

The sample to be examined consisted of clusters of 250 persons, in 48 sample villages with a total of some 12,000 persons. Since a meaningful stratification of the population to be examined was not possible, 48 villages out of the 1,605 villages in the whole survey area were selected, with probabilities proportional to their population size. The first village was chosen at random in the first 48th fraction of the total population. A further 47 villages were determined by periodical sampling at similar intervals. When comparing the seroreactor rates found in the 48 villages at ITS with the seroreactor rates of the administrative units (MNOs) to which the villages belonged, the 48 selected villages were found to form a good representative sample of he areas from which they were selected.

Table IV (opposite) lists the villages with the clinical and serological findings at ITS in the villages and the corresponding MNOs. Fig. 4 shows the geographical distribution of the selected villages.

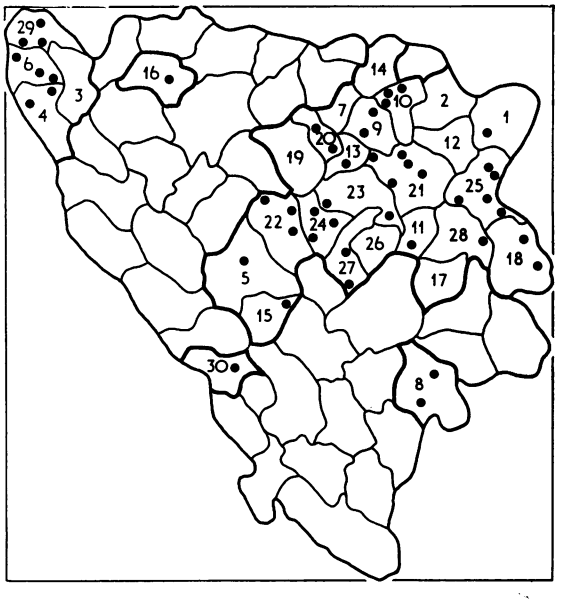

FIG. 4 *Geographical distribution of the* 48 *selected villages*

A satisfactory correlation between reactor-rates at ITS in the selected villages and the corresponding MNOs was found; this is shown in Fig. 5 (overleaf).

*The following subjects were studied in the multipurpose investigations of the Bosnian sera:
(1) *Tetanus immunity* Gamaleya Institute of Epidemiology and Microbiology, Moscow (1,053 sera from six sample villages).
(2) *Antistreptolysin-0, diphtheria, pertussis, and parapertussis* Institute of Epidemiology and Microbiology, Prague (591 sera from selected age groups from eight sample villages).
(3) *Arbovirus infections* Department of Virology, School of Public Health, University of Zagreb (2,115 sera from fourteen sample villages).
(4) *Rickettsial infections* Department of Epidemiology, Medical Faculty, Sarajevo (1,041 sera from six sample villages).
(5) *Malaria* Nuffield Institute for Medical Research, London (300 sera from four sample villages).
(6) *Human genetic factors* Centre Départmental de Transfusion Sanguine, Rouen (760 sera from four sample villages).
The reports on these investigations are to be published separately.

TABLE IV *Basic findings at ITS in the selected villages and the corresponding MNOs*

Selected village no.	*Srez code number*	*MNO out of which village was selected*				*Selected village*		
		Code number	*Name*	*Rate of infective lesions (per cent.)*	*Sero-reactor rate (per cent.)*	*Code number*	*Name*	*Sero-reactor rate (per cent.)*
1	01	04	Ugljevik	0·29	2·2	07	Dragovići	2·8
2	04	01	Vrsta	0·05	5·6	08	Mala Peča	5·6
3	04	02	Pokoj	0·05	2·1	02	Srbljani	2·5
4	05	06	Donji Vakuf	0·02	1·5	04	Sandzak etc.	1·9
5	06	02	Liskovac	0·28	9·0	02	Čajici	6·7
6	20	04	Piljozići	0·61	6·2	02	Lončari	0·60
7	20	13	Stijena	1·1	9·4	01	Podgredina	10·9
8	08	03	Ustikolina	0·11	0·92	03	Paunči	0·0
9	08	06	Sutješka	0·04	0·62	02	Djedjevo	0·40
10	09	03	Petrovo Selo	0·00	1·3	04	Krtova	2·1
11	09	07	Sladna	4·6	25·0	05	Pirage	25·7
12	10	03	Zelinja Gornja	1·7	22·8	02	Jasenice	17·5
13	10	10	Vida	0·37	19·1	01	Novalići	33·0
14	10	11	Ledenice	0·07	7·2	03	Led. Muslimanske	8·2
15	11	06	Olovo	0·20	6·4	02	Boganovići	4·8
16	13	—	—	0·19	2·9	14	Ulišnjak Donji	0·94
17	15	—	—	0·02	0·98	16	Scipe	0·95
18	20	07	Jelah	0·00	1·7	05	Vukovo	1·5
19	18	02	Srebrenica	0·11	2·6	04	Dimnići etc.	2·8
20	18	03	Bratunac	0·06	2·4	07	Bljeceva	0·28
21	20	07	Jelah	0·07	1·6	02	Jevadzije	2·6
22	20	06	Kalosevic	0·57	5·3	02	Kalosevic	4·8
23	21	05	Banovici	1·0	7·9	02	Trestenica etc.	11·6
24	21	08	Tojsic	0·77	7·3	07	Gornja Vukuvija	2·6
25	21	11	Poljana	0·15	4·5	07	Bistarac D.G.	2·0
26	21	16	Tinja Brezik	0·47	6·8	07	Podpeč	0·88
27	21	20	Vasiljevći	0·00	0·00	03	Vasiljevći	0·00
28	22	02	Vitez	0·07	0·96	04	Sadovaća etc.	1·1
29	22	04	Guća Gora	0·02	2·2	03	Višnjevo	10·0
30	22	05	Turbe	0·00	0·92	11	Varošluk	0·4
31	23	01	Golubinja	0·62	4·0	05	Mestova-Ravna	7·5
32	23	02	Careva Cuprija	0·42	13·9	03	Rijeka	12·2
33	24	09	Nemila	0·00	3·8	01	Nemila	4·7
34	24	15	Brnjić	0·00	3·5	05	Krsevać	0·00
35	24	16	Cajdras	0·00	6·7	01	Visokovći	3·2
36	25	01	Drinjaća	0·12	5·7	05	Lijesanj	1·4
37	25	02	Osmaci	0·25	10·5	09	Jelovo Brdo	30·6[a]
38	25	05	Kozluk	0·12	13·4	01	Tabanči	5·9
39	25	08	Grbavći	0·69	12·0	04	Krisevići	10·6
40	25	15	Čalopek	0·63	17·0	01	Tršić	15·8
41	27	01	Gračanica	0·00	4·2	09	Uvorići	5·8
42	27	04	Kraljeva Sutj.	0·00	4·7	05	Teševo	1·7
43	27	09	Podgora	0·13	1·7	04	Ocevlje etc.	2·0
44	28	07	Milići	0·00	2·3	06	Rajiči etc.	0·60
45	29	01	Todorovc	0·36	1·9	05	Vejinac	0·49
46	29	03	Mala Kladuša	0·02	2·7	01	Glavica	4·3
47	29	04	Velika Kladuša	0·08	3·9	07	Kumarica	7·9
48	30	—	—	0·00	1·1	03	Poklecani	0·28

[a]Administrative division at the time of the ITS

The average seroreactor rate in the population examined at the ITS was 5·9 per cent. (Table II). The corresponding estimated rate which would have been obtained from the examination of a cluster of 250 persons in the selected villages in the present sample is 6·4 per cent., with an approximate 95 per cent. confidence interval ranging from 5·2 to 7·6 per cent. The sample is therefore considered to be reasonably representative of the population in this respect.

In each of the 48 sample villages, a population cluster comprising 250 persons was selected on a household basis by survey teams in the field. For this purpose up-to-date census lists were established for the entire village and a random sample was selected by means of a list of random numbers.

Field procedures

The field survey procedures included the use of household cards which were completed during the village census. An individual record card was prepared for each person in the selected cluster. For the retrospective study a special individual card was

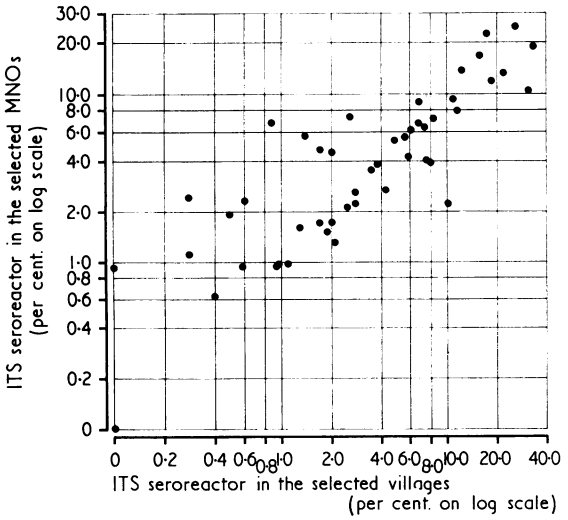

FIG. 5 *Correlation between the reactor rates at ITS in the selected villages and the corresponding MNOs*

established for recording data on individuals whom personal, clinical, serological, and other information was available from the previous mass campaign in the central registry in Sarajevo. All persons selected in the clusters were checked against this register.

The field procedures comprised history taking, clinical inspection, venepuncture, and preparation of dried blood specimens (rondelles). Arrangements for optimal transport of specimens were made. Deep-frozen specimens were shipped to WHO reference centres and the collaborating laboratories, as specified in the Survey Design and Working Protocol (INT/VDT/70.315).

The field procedures included, finally, provision of certain curative medical care for simple illnesses among the village populations.

Laboratory procedures

All sera were examined at the Sarajevo laboratory by the VDRL, Meinecke, and FTA tests. A subsample, representing all sera obtained in twelve out of 48 villages, was examined by the TPI test also. These sera were examined in the same tests at the WHO Reference Centre in Copenhagen. A special 'triple assay' or proficiency study of the TPI test results was organized between the Reference Centres in Copenhagen and Atlanta and the Sarajevo laboratory.

The deep-frozen serum aliquots in Sarajevo were also shipped in liquid nitrogen to several collaborating laboratories for immunological tests for various other conditions.

Data processing

All data were collected at WHO headquarters for collation and exploitation in co-operation with the Institute in Sarajevo, the WHO reference centres, and the collaborating laboratories.

Results of the Study

The results of the general seroepidemiological study carried out from 1968 to 1970 will be presented first,

TABLE V *Coverage of the selected sample, according to age groups and sex*

Age group (yrs)	*Male*				*Female*				*Both sexes*			
	Selected	*Absent*[a]	*Examined*	*Per cent.*	*Selected*	*Absent*[a]	*Examined*	*Per cent.*	*Selected*	*Absent*[a]	*Examined*	*Per cent.*
0–1	231	154	77	33·3	241	170	71	29·5	472	324	148	31·4
2–4	527	106	421	79·9	515	105	410	79·6	1,042	211	831	79·8
5–9	965	94	871	90·3	897	75	822	91·6	1,862	169	1,693	90·9
10–14	927	87	840	90·6	860	83	777	90·3	1,787	170	1,617	90·5
0–14	2,650	441	2,209	83·4	2,513	433	2,080	82·8	5,163	874	4,289	83·1
15–29	1,263	392	871	69·0	1,504	184	1,320	87·8	2,767	576	2,191	79·2
30–44	1,054	434	620	58·8	1,212	140	1,072	88·4	2,266	574	1,692	74·7
45–59	537	143	394	73·4	650	92	558	85·8	1,187	235	952	80·2
60+	368	81	287	78·0	357	87	270	75·6	725	168	557	76·8
15+	3,222	1,050	2,172	67·4	3,723	503	3,220	86·5	6,945	1,553	5,392	77·6
Total	5,872	1,491	4,381	74·6	6,236	936	5,300	85·0	12,108	2,427	9,681	80·0

[a]Absent comprises selected persons who either did not present themselves or refused examination as well as individuals from whom no adequate serum sample could be obtained for serological examination

to be followed by the results of the retrospective study based on the records from the mass campaign.

General seroepidemiological study

(i) Sampling performance

A total sample population of 12,108 persons was selected in 48 clusters. Table V shows the coverage of the selected sample according to age-groups and sex. The average coverage was 80 per cent. Most of the missed coverage was caused by failure to obtain sufficient blood for serological examination from the youngest children of both sexes, either because the parents refused to submit their children to blood-letting or because of failure of technique. A second cause was absence from the examination of adult men, many of whom worked at a considerable distance from their villages and, in many instances, came home late in the evening or during weekends only. Notwithstanding the 20 per cent. loss of coverage, the population sample remains sufficiently representative to allow conclusions to be drawn.

TABLE VI *Seroreactor rates (Meinicke test) at ITS and as established in seroepidemiological evaluation study, according to sample village*

Survey no.	*Village name*	*ITS seroreactor rate*	*Sero-epidemiological study 1968 seroreactor rate*	*Population by nationality (per cent.)*		
				Muslims	*Serbs*	*Croats*
1	Dragovici	2·8	1·8	0·4	99·6	—
2	Mala Peca	5·6	3·0	100·0	—	—
3	Srbljani	2·5	1·8	100·0	—	—
4	Sandzak etc.	1·9	1·5	56·5	43·5	—
5	Cajici	6·7	0·6	100·0	—	—
6	Loncari	0·60	0·0	—	2·4	97·6
7	Podgredina	10·9	0·0	100·0	—	—
8	Paunci	0·0	0·0	95·6	4·4	—
9	Djedjevo	0·40	0·0	61·1	38·9	—
10	Krtova	2·1	0·0	—	100·0	—
11	Pirage	25·7	3·1	98·7	1·3	—
12	Jasenice	17·5	4·1	88·2	11·8	—
13	Novalici	33·0	2·3	100·0	—	—
14	Led. Muslimanske	8·2	2·6	100·0	—	—
15	Boganovici	4·9	4·8	100·0	—	—
16	Ulisnjak Donji	0·94	0·0	100·0	—	—
17	Scipo	0·95	0·0	79·5	—	20·5
18	Vckovo	1·5	0·4	100·0	—	—
19	Dimnici etc.	2·8	5·1	100·0	—	—
20	Bljeceva	0·28	0·5	95·4	4·6	—
21	Yevadia	1·3	2·6	91·8	5·2	3·0
22	Kalosevic	4·8	0·0	100·0	—	—
23	Trestenica etc.	11·6	1·8	27·0	73·0	—
24	Gornja Vukuvija	2·6	0·5	100·0	—	—
25	Bistarac D.G.	2·0	1·2	50·0	—	50·0
26	Podpec	0·88	1·4	0·4	99·6	—
27	Vasiljevci	0·0	0·0	—	100·0	—
28	Sadovaca etc.	1·1	1·0	89·2	—	10·8
29	Visnjevo	10·0	2·5	100·0	—	—
30	Varosluk	0·58	0·0	—	100·0	—
31	Nesto Ravan	7·5	1·0	100·0	—	—
32	Rijeka	12·2	1·7	87·8	2·4	9·8
33	Nemila	4·7	2·4	100·0	—	—
34	Bistrica	0·0	0·0	32·8	67·2	—
35	Visokovci	3·2	0·0	62·0	—	38·0
36	Lijesanj	1·4	0·0	0·8	99·2	—
37	Jelovo Brdo	30·6	2·3	89·3	10·7	—
38	Tabanci	5·9	0·0	0·0	100·0	—
39	Krisevici	10·6	1·6	100·0	—	—
40	Trsic	15·8	0·5	77·3	22·7	—
41	Uvorici	5·8	3·9	96·5	3·5	—
42	Tesevo	1·7	0·0	—	100·0	—
43	Ocevlje etc.	2·0	0·5	27·6	72·4	—
44	Rajici etc.	0·60	0·4	0·8	99·2	—
45	Vejinac	0·49	0·0	100·0	—	—
46	Glavica	4·3	1·2	100·0	—	—
47	Kumarica	7·9	1·6	100·0	—	—
48	Foklecani	0·28	0·0	0·4	—	99·0
Total sample				67·0	26·2	6·7

(ii) Results of serological testing in the Sarajevo laboratory

Table VI shows the average seroreactor rates at the ITS in the 48 sample villages, compared with the rates during the present seroepidemiological study, based on the Meinicke test, the only test used on both occasions. A considerable decrease is noted in all the sample villages except three. In many villages no seroreactors were found at all. The average initial rate at ITS was shown to be 5·94 per cent. (Table II). In the sample population examined in 1968 the average seroreactivity rate in the same test was 1·22 per cent. (Table VII)—a considerable decrease. The previous socioeconomic conditions of the Muslim ethnic group have been shown to have resulted in a higher prevalence of infection than in other groups and higher in females than in males. The seroreactor rates found in the evaluation study in 1968 were therefore broken down according to ethnic groups in Table VII. The reactor rate in the Muslim population was still higher than in the other groups, but in all groups the rates had decreased significantly.

It should be noted that the impact of the mass campaign coincided with an impressive improvement of the socioeconomic conditions throughout Yugoslavia after the war, and of the peasant Muslim population of Bosnia in particular. The mass campaign lasted from 1948 to 1955. The children up to 15 years old examined in the evaluation study in 1968 were all born after the mass campaign and grew up in a period of rapid environmental improvement, *e.g.* industrialization, schooling, improvement of communications, intensive medical and social welfare activity, etc. Clinical lesions of endemic syphilis having disappeared from Bosnia, the investigation of seroreactivity in the children was therefore of utmost importance to evaluate the effect of the mass campaign. As the TPI test is the most specific indicator of past and—to some extent—recent treponemal infection, the TPI results will be discussed first. 2,358 sera were tested from a subsample of twelve villages which had a much higher rate of endemic syphilis than the average in the endemic area at the time of the mass campaign. The results (Table VIII) indicate the absence of TPI reactors in the youngest children (up to 5 years old). One TPI seroreactor (a boy aged 9 years) was found in the next age group of 5 to 9 years. The next older group of children, 10 to 14 years, showed no seroreactors. The adults, from 15 years onwards, showed an age-related increase in seroreactivity, the maximum being 13·3 per cent in the 45 to 59-year age group.

The TPI test results differ from results of the FTA-200 and Meinicke tests (Table VIII), but there is considerable agreement with the VDRL rates in the different age groups. In view of the high specificity of the TPI test, the much higher seroreactivity rates obtained by FTA-200 testing suggest a high proportion of false reactive FTA-200 results. Table IX indicates that fifty out of 95 reactive FTA-200 results were non-reactive in the TPI test. The sensitivity of the FTA-200 test was also unsatisfactory when compared with the TPI test: only 45 (69 per cent.) of 65 TPI-reactive sera were found to be reactive in the FTA-200 test. On the other hand, the results of the VDRL test are closer to the TPI test results. The

TABLE VII *Seroreactor rates in Meinicke and VDRL tests, according to nationality in two age groups (Sarajevo laboratory)*

Nationality	*Age group (yrs)*	*Meinicke test*			*VDRL test*		
		No. examined	*Reactive*		*No. examined*	*Reactive*	
			No.	*Per cent.*		*No.*	*Per cent.*
Muslims	0–14	2,715	4	0·15	2,714	4	0·15
	15+	3,526	101	2·86	3,527	266	7·54
	Total	6,241	105	1·68	6,241	270	4·33
Serbs	0–14	1,013	1	0·10	1,013	0	0·00
	15+	1,507	8	0·53	1,507	14	0·93
	Total	2,520	9	0·36	2,520	14	0·56
Croats	0–14	229	0	0·00	229	0	0·00
	15+	335	0	0·00	334	6	1·80
	Total	564	0	0·00	563	6	1·07
Total	0–14	3,957	5	0·13	3,956	4	0·10
	15+	5,368	109	2·03	5,368	286	5·33
	Total	9,325	114	1·22	9,324	290	3·11

TABLE VIII *Test results in TPI, FTA-200, VDRL, and Meinicke tests, according to age (Sarajevo laboratory)*

	TPI				*FTA-200*			*VDRL*				*Meinicke*			
Age group (yrs)	*Tested*	±	+	*Total reactive per cent.*	*Tested*	+	*per cent.*	*Tested*	±	+	*Total reactive per cent.*	*Tested*	±	+	*Total reactive per cent.*
0–1	8	0	0	0·0	30	0	0·0	30	0	0	0·0	30	0	0	0·0
2–4	183	0	0	0·0	669	4	0·6	670	1	1	0·3	670	2	0	0·3
5–9	458	1	0	0·2	1,650	5	0·3	1,652	0	0	0·0	1,652	1	0	0·1
10–14	386	0	0	0·0	1,603	24	1·5	1,604	2	0	0·1	1,605	2	0	0·1
0–14	1,035	1	0	0·1	3,952	33	0·8	3,956	3	1	0·1	3,957	5	0	0·1
15–29	573	1	2	0·5	2,178	36	1·7	2,184	8	7	0·7	2,184	8	4	0·5
30–44	409	4	14	4·4	1,681	111	6·6	1,682	35	28	3·7	1,682	20	5	1·5
45–59	218	13	16	13·3	948	141	14·9	948	85	30	12·1	947	29	6	3·7
60+	123	9	6	12·2	553	106	19·2	554	50	43	16·8	555	22	15	6·7
15+	1,323	27	38	4·9	5,360	394	7·4	5,368	178	108	5·3	5,368	79	30	2·0
Total	2,358	28	38	2·8	9,312	427	4·6	9,324	181	109	3·1	9,325	84	30	1·2

TABLE IX *Comparison of TPI and FTA-200 results (Sarajevo laboratory)*

FTA Sarajevo	*TPI—Sarajevo*			
	−	±	+	Total
−	2,240	8	12	2,260
+	50	20	25	95
Total	2,290	28	37	2,355

Co-negativity: 2,240/2,290 = 97·8 per cent.
Co-positivity: 45/65 = 69·2 per cent.
Overall agreement: 2,285/2,355 = 97·0 per cent.

Meinicke results in Sarajevo show much lower rates of reactivity than the other serological tests; it was included in the battery of tests used, as it had been used extensively during the previous mass campaign.

(iii) Serological results at the WHO Reference Laboratory, Copenhagen

Table X (overleaf, pp. 12 and 13) shows the results obtained in the treponemal antigen tests (TPI and FTA-200) and in the reagin tests (VDRL, Meinicke, Wassermann, and Kahn) in the Reference Laboratory. TPI-reactors were found in the age groups from 2 to 4 years onwards. The reactivity rates were low up to 10 to 14 years and thereafter increased with age. FTA-200 and VDRL rates are consistently higher in all age groups than the TPI rate.

(iv) Comparison of TPI and FTA-200 results at different laboratories

Comparison of specificity and sensitivity of the TPI and FTA-200 results between the two laboratories is shown in Tables XI and XII. Apparently the sensitivity of both tests was greater in the Reference Laboratory than in Sarajevo. To study the differences in the results with particular reference to the TPI test, a blind 'triple assay' was organized on the basis of a collection of selected sera comprising most of

TABLE XI *Comparison of TPI test results in Sarajevo and Copenhagen*

TPI Copenhagen	*TPI Sarajevo*			
	−	±	+	Total
−	2,142	17	16	2,175
±	27	6	2	35
+	29	4	19	52
Total	2,198	27	37	2,262

Co-negativity: 2,142/2,175 = 98·5 per cent.
Co-positivity: 31/87 = 35·6 per cent.
Overall agreement: 2,173/2,262 = 96·1 per cent.

TABLE XII *Comparison of FTA-200 test results in Sarajevo and Copenhagen*

FTA Copenhagen	*FTA Sarajevo*		
	−	+	Total
−	2,042	28	2,070
+	143	68	211
Total	2,185	96	2,281

Co-negativity: 2,042/2,070 = 98·6 per cent.
Co-positivity: 68/211 = 32·2 per cent.
Overall agreement: 2,110/2,281 = 92.5 per cent.

TABLE X *Test results in TPI, FTA-200, VDRL, Meinicke tests, Wassermann reaction, and Kahn tests according to age*

Age group (yrs.)	TPI				FTA-200			VDRL			
	Tested	±	+	Total reactive per cent.	Tested	+	Per cent.	Tested	±	+	Total reactive per cent.
0–1	6	0	0	0·0	6	0	0·0	6	1	0	16·7
2–4	148	1	2	2·0	150	5	3·3	146	8	0	5·5
5–9	430	4	3	1·6	433	11	2·5	419	11	1	2·9
10–14	381	3	3	1·6	381	19	5·0	369	13	0	3·5
0–14	965	8	8	1·7	970	35	3·6	940	33	1	3·6
15–29	564	9	12	3·7	569	38	6·7	552	24	4	5·1
30–44	409	5	13	4·4	408	62	15·2	404	31	23	13·4
45–59	214	9	12	9·8	217	39	18·0	208	26	27	25·5
60+	122	6	8	11·5	122	38	31·1	118	14	22	30·5
15+	1,309	29	45	5·7	1,316	177	13·4	1,282	95	76	13·3
Total	2,274	37	53	4·0	2,286	212	9·3	2,222	128	77	9·2

those found reactive in either Copenhagen or in Sarajevo (or both). The triple assay concerned 293 coded serum specimens and testing was carried out in the Sarajevo and Copenhagen laboratories, as well as at the VDRL laboratory, USPHS, Atlanta.

(v) *Reproducibility of the TPI test*

The reproducibility of the TPI test has frequently been studied, especially during the first years after its introduction at the WHO International Treponematoses Laboratory Center, Johns Hopkins University, in 1949. Interlaboratory investigations undertaken through WHO were unable to verify the limits within which the test could be reproduced in different laboratories. In the context of the present study of 'disappearing' endemic treponematoses in Bosnia, it was considered necessary to examine these aspects further. Moreover, proficiency testing is being undertaken in a number of leading centres, the results of which will be published in the near future. The data obtained in the triple assay regarding sera from the study in Bosnia are voluminous and will require extensive statistical analysis, the results of which will be published separately. A preliminary study has, nevertheless, shown that some inter-laboratory variation occurs. Intra-laboratory variation in the results of testing of paired, coded, sera was also noted. Both the inter- and the intra-laboratory variations occurred mainly with weakly reactive sera which are sometimes diagnosed as non-reactive and sometimes as reactive. The testing of clearly reactive or non-reactive sera rarely gives rise to discrepancies. Before extensive analysis of carefully designed proficiency testing becomes available, it is recommended that some caution should be exercised in the interpretation of the results of TPI testing. For the vast majority of TPI tests carried out, unequivocal conclusions are justified, but in a minority of cases it appears prudent not to consider the TPI test results as the final and absolute criterion of past or present treponemal infection, but to take all other clinical and serological evidence into account.

Preliminary analysis of the triple assay data showed that intra-laboratory variation in results of blind testing of paired sera did not occur in the Sarajevo laboratory. Furthermore, it was shown that a remarkable consistency existed in the Sarajevo laboratory results of TPI tests of coded sera obtained from the same individual at different periods. It is therefore considered appropriate to use the results of the TPI testing in the Sarajevo laboratory to evaluate the outcome of the mass campaign, especially as almost all of the Sarajevo results were confirmed by the findings in the Reference Laboratories. Discrepancies between test results in children born after the mass campaign were extremely rare, and taking into account the non-reactivity of these sera in almost all the other serological tests used these should be regarded as open to varying interpretations.

The analysis of the age-specific TPI-reactivity rates (Table VIII) brought to light only one TPI-reactive individual (a 9-year-old boy) among the children under 14 years of age, that is among those born after the completion of the mass campaign in 1953. For the last 8 years no child was shown to have

(*Copenhagen laboratory*)

einicke				*Wassermann*			*Kahn*		
ted	±	+	*Total reactive per cent.*	*Tested*	+	*Total reactive per cent.*	*Tested*	+	*Total reactive per cent.*
6	1	0	16·7	5	0	0·0	2	0	0·0
123	1	0	0·8	143	0	0·0	120	3	2·5
359	12	0	3·3	428	1	0·2	405	2	0·5
324	17	0	5·2	374	0	0·0	366	5	1·4
812	31	0	3·8	950	1	0·1	893	10	1·1
478	24	1	5·2	568	0	0·0	554	8	1·4
343	15	2	5·0	403	2	0·5	390	3	0·8
182	18	0	9·9	212	1	0·5	201	5	2·5
104	16	1	16·3	119	1	0·8	106	4	3·8
1,107	73	4	7·0	1,302	4	0·3	1,251	20	1·6
1,919	104	4	5·6	2,252	5	0·2	2,144	30	1·4

been infected. Together with the absence of any active clinical cases, this sero-negativity permits us to conclude that complete eradication, according to the criteria of the WHO Expert Committee of 1961, was attained.

Retrospective seroepidemiological study

During the mass campaign against endemic treponematosis in Bosnia, individual records were established of all the findings during the ITS and the consecutive RSs. The preservation of the records concerning individuals examined at that time made it possible to assess the outcome of the mass campaign by comparing the findings in the present seroepidemiological study with those concerning the same individuals at the time of the mass campaign (Tables XIV to XXIV).

The children born after the campaign and examined in 1968 had, of course, no previous records. This group accounts for 4,289 individuals out of a total of 9,681 examined in the seroepidemiological evaluation study (Table V). On the other hand, many individuals examined in the mass campaign (1948–55) died before 1968, and their previous records could not be used for comparison with the 1968 data. In the years which have elapsed since the beginning of the mass campaign, from 15 to 20 per cent. of the population have thus disappeared. Therefore, of 5,392 persons over 15 years of age examined in 1968, mass campaign records could be expected to be available for approximately only 80 to 85 per cent. (4,000 to 4,500 individuals). In fact, ITS records concerning 2,402 persons were retrieved from the Central Registry at Sarajevo, representing 55 to 60 per cent. of the highest possible number. Regarding

TABLE XIII *Comparison of the distribution by sex and nationality of (A) the persons for whom ITS records were retrieved, and (B) the total sample population examined in* 1968

Group	*Muslims*			*Serbs*			*Croats*			*Total*		
	Male	*Female*	*Total*	*Male*	*Female*	*Total*	*Male*	*Female*	*Total*	*Male*	*Female*	*Total*
Number examined in 1968	2,811	3,430	6,241	1,132	1,388	2,520	255	309	564	4,198	5,127	9,325
Number with ITS records	821	771	1,592	350	346	696	54	60	114	1,225	1,177	2,402
Percentage with ITS records	29·2	22·5	25·5	30·9	24·9	27·6	21·2	19·4	20·2	29·2	23·0	25·8

the 1,500 to 2,000 individuals for whom no previous records could be retrieved, it should be noted that—in the course of the last 20 years—a great number of families and individuals moved to other villages and towns (urbanization) and could not be located in 1968 in the 48 sample villages where they had lived at the time of the mass campaign.

The examination of the background data on 2,402 individuals permits retrospective evaluation of the mass campaign from another angle than that of the general assessment of the seroepidemiological findings as presented above.

Table XIII shows that the distribution of individuals according to sex and nationality was approximately the same in the retrieved ITS records as in the sample examined during the seroepidemiological study. It may, therefore, be assumed that the 2,402 mass campaign records available represent an acceptable sample of the Bosnian population.

TABLE XIV *Persons examined at ITS and subsequent re-surveys of the mass campaign during the seroepidemiological study* (1968)

Calendar year	*Examined at ITS*	*1st RS*	*2nd RS*	*3rd RS*	*4th RS*
1948	206	—	—	—	—
1949	538	—	—	—	—
1950	300	103	—	—	—
1951	519	339	103	—	—
1952	347	388	64	—	—
1953	347	153	412	103	—
1954	98	209	49	—	—
1955	—	—	—	—	—
1956	47	—	—	—	—
1957	—	—	—	—	—
1958	—	—	—	—	—
1959	—	—	—	—	—
1960	—	—	—	—	—
1961	—	—	—	—	103
Total	2,402	1,192	628	103	103

The numbers of individuals examined at subsequent RSs after ITS decreased rapidly at each successive RS (Table XIV). Table XV shows that at ITS the policy was treatment of all overt active and inactive clinical cases and latent cases (seroreactors). The number of contacts treated was limited—only 22 out of a total of 2,370 persons (0·93 per cent.). At ITS seven cases of infectious lesions were found in 2,370 persons examined (ITS data were retrieved for 32 persons who were younger than 15 years in 1968 but none had a record of clinical or serological findings and they are therefore not considered here). This is 0·3 per cent. as compared to an average of 0·44 per cent. in the whole population observed at ITS (Table IIa). The much higher prevalence of clinical and serological endemic syphilis in the Muslim group than in the other groups is also shown in Table XV.

Table XVI shows a characteristic hypoendemic area with benign infectious lesions in some age groups. The infectious lesions in persons concerning whom ITS data were available were found mainly among the adolescent and adult groups. The same appears to apply to non-infectious and inactive lesions, as far as the small numbers allow conclusions. The majority of seroreactors were also found in the higher age groups.

The clinical cases shown in Table XVII were found in eight villages only. At ITS five infectious cases were detected in Village 31 and two in Village 47, where also one active non-infectious case was found. The six inactive cases were found in Villages 1, 3, 11, 32, 37, and 46.

TABLE XV *Clinical and serological findings at ITS of 2,370 persons aged 15 years or older treated as case or contact or not treated at ITS, according to nationality*

Diagnosis at ITS	*Muslims*				*Serbs*				*Croats*				*Total*			
	Case treatment	*Contact treatment*	*Not treated*	*Total*	*Case treatment*	*Contact treatment*	*Not treated*	*Total*	*Case treatment*	*Contact treatment*	*Not treated*	*Total*	*Case treatment*	*Contact treatment*	*Not treated*	*Total*
Active infectious lesions	7	0	0	7	0	0	0	0	0	0	0	0	7	0	0	7
Active non-infectious lesions	1	0	0	1	0	0	0	0	0	0	0	0	1	0	0	1
Inactive lesions	5	0	0	5	1	0	0	1	0	0	0	0	6	0	0	6
No lesions but seroreactive	94	0	0	94	14	0	0	14	0	0	0	0	108	0	0	108
Negative clinical and serological findings	0	21	1,433	1,454	0	1	679	680	0	0	114	114	0	22	2,226	2,248
Total	107	21	1,433	1,561	15	1	679	695	0	0	114	114	122	22	2,226	2,370

122 clinical or serological cases treated, 22 contacts treated, 2,326 persons untreated

TABLE XVI *Distribution of 122 persons treated at ITS as clinical or serological* cases, according to age at ITS*

Age at ITS (yrs)	*Infectious lesions*	*Active non-infectious cases*	*Inactive lesions*	*Clinically negative*		*Total*
				Serology reactive	*Serology weakly reactive*	
0–4	—	—	—	1	—	1
5–9	—	—	—	2	2	4
10–14	1	—	2	8	—	11
15–29	2	—	—	20	9	31
30–44	4	1	2	36	9	52
45–59	—	—	2	14	7	23
60+	—	—	—	—	—	—
Total	7	1	6	81	27	122

*Meinicke test

During the first RS, one active infectious case was recorded in Village 12, together with one active but non-infectious case. No further clinical cases are to be found in the available records of the mass campaign. It is remarkable that the villages where the seven ITS infectious cases were found did not belong to the most heavily infected villages (Table VI).

TABLE XVII *PAM treatment schedules used at ITS in the 122 active cases*

Number of injections	*Total dosage of PAM (ml.)*[a]											*Total cases treated*
	6	7	8	9	10	11	12	13	14	15	16	
1	1	—	2	—	1	—	11	—	—	—	—	15
2	—	—	3	—	2	—	22	—	—	—	—	27
3	—	—	—	—	—	—	20	—	—	—	—	20
4	—	—	—	—	1	—	0	—	—	—	1	2
5	—	—	—	—	1	—	1	—	—	—	—	2
6	—	—	—	—	0	—	11	—	1	—	—	12
7	—	—	—	—	1	—	0	—	43	—	—	44
Total	1	0	5	0	6	0	65	0	44	0	1	122

[a]1 ml. = 0·3 m.u.
Duration of treatment: 69 cases treated in 1 wk; 46 cases treated in 2 wks; 7 cases treated in 3 wks

TABLE XVIII *Treatment at initial survey (ITS) and at subsequent re-surveys (RS) of persons for whom records from mass campaign were available*

Decision for treatment	*ITS*		*1st RS*		*2nd RS*		*3rd RS*		*4th RS*	
	Number	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*
Case (clinical or serological)	122	5·1	25	2·1	12	1·9	0	0	6	5·8
Contact	22	0·9	1	0·1	1	0·2	0	0	0	0
No treatment	2,258	94·0	1,166	97·8	615	97·9	103	100	97	94·2
Total	2,402	100	1,192	100	628	100	103	100	103	100

Table XVII gives the dosage schedule of long-acting penicillin (PAM) used for case treatment: the great majority of cases received 12 or 14 ml. (3·6 to 4·2 m.u.) in a varying number of injections, usually over 1 to 2 weeks. In only seven cases did the course of treatment last 3 weeks. Table XVIII shows that the rate of cases treated decreased during subsequent surveys and that contact treatment, already limited at ITS, was applied even less frequently during the RSs.

The results of serological testing at ITS and subsequent RSs did not show an impressive decrease of reactor rate as the mass campaign progressed (Table XIX). This should be seen in the light of the observation that in hypoendemic areas, like Bosnia, initially low reactor rates do not drop dramatically, as in hyperendemic areas. Also, the careful selection of people to be examined at RSs (which resulted in rapidly decreasing population coverage on consecutive occasions) increased the relative importance of reactors at later RSs, which made it seem that there was an increase in the number of reactors. More revealing are the results of serological testing in consecutive RSs of clinical and serological cases detected at ITS (Table XX). It is noted that the reactor rates decreased rapidly after treatment during ITS, as shown by the test results at the first and second RSs. (The apparent increase in seroreactivity in later RSs is due to an increased relative importance of reactors in the selective and continuously decreasing number of tests carried out.) The effectiveness of the penicillin treatment is thus borne out and the clinical rate at ITS of 0·3 per cent. decreased to 0·07 per cent. at the first RS and to nil at the second RS.

The results of the Meinicke test in 1968 in the same 122 individuals treated as cases during ITS are shown in Table XXI. The majority of reactors at ITS were found to be non-reactive in 1968 (64 out of 94). Only six (4·9 per cent.) were still reactive, the remainder being weakly reactive. Of those weakly reactive at ITS, the majority were non-reactive in 1968. Two were found to be reactive, which may be

TABLE XXI *Comparison of results of Meinicke tests during ITS and during seroepidemiological study in* 122 *persons who had been treated as clinical or serological cases at ITS*

Meinicke test ITS	*Meinicke test, 1968*				
	+	±	—	*Unknown*	*Total*
+	6	24	64	1	95
±	2	2	17	1	22
—	0	0	4	0	4
Not done	0	0	1	0	1
Total	8	26	86	2	122

TABLE XIX *Results of Meinicke test at ITS and subsequent re-surveys*

Meinicke test results	*ITS*		*1st RS*		*2nd RS*		*3rd RS*		*4th RS*	
	Number	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*
+	96	4·7	19	2·1	10	1·8	2	3·5	4	4·3
±	40	1·9	28	3·1	15	2·8	1	1·8	8	8·5
—	1,921	93·4	846	94·7	517	95·4	54	94·7	82	87·2
Total	2,057	100	893	100	542	100	57	100	94	100

TABLE XX *Meinicke results at ITS and subsequent re-surveys of* 122 *clinical cases or latent cases found and treated at ITS*

Meinicke test results	*ITS*		*1st RS*		*2nd RS*		*3rd RS*		*4th RS*	
	Number	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*
Positive	95	78·5	12	15·4	8	12·7	1	16·7	3	50
Weakly positive	22	18·2	14	18·0	9	14·3	1	16·7	2	33·3
Negative	4	3·3	52	66·7	46	73·0	4	66·6	1	16·7
Examined	121	100	78	100	63	100	6	100	6	100
Not done	1		44		59		116		116	
Total	122		122		122		122		122	

explained by differences in test techniques so many years later. The individuals who remained reactive or weakly reactive up to 1968, all adequately treated at ITS and without signs of clinical lesions, must be considered to have residual antibody. In four persons who were seronegative both at ITS and in 1968, the diagnosis at ITS was not precisely stated.

Table XXII shows the findings in the Meinicke test of persons left untreated at ITS. Out of 1,897 individuals tested, five were reactive in 1968 and nineteen weakly reactive. Among these 24 reactors, one only was weakly reactive at ITS. Differences, such as antigens used, sensitivity of test, and technique used some 20 years after ITS, may explain this observation, in the absence of recognized active clinical cases of endemic syphilis in Bosnia since 1959.

TABLE XXII *Results of Meinicke testing during ITS and seroepidemiological study of* 1,897 *persons who were not treated at ITS*

Meinicke test ITS	*Meinicke test*, 1968			
	+	±	−	Total
+	0	0	1	1
±	1	0	17	18
−	4	19	1,855	1,878
Total	5	19	1,873	1,897

TABLE XXIII *Meinicke results* (1968) *in* 120 *clinical or serological cases found at ITS, according to age*

Age group (yrs)	*Meinicke test*, 1968				
	No. tested	±	+	−	*Per cent. reactive*
15–29	8	1	0	7	12·5
30–44	30	7	1	22	26·7
45–59	53	10	2	41	22·6
60+	29	8	5	16	44·8
Total	120	26	8	86	28·3

Table XXIII shows the distribution of the 1968 Meinicke test results of 120 cases found at ITS, according to age. As would be expected, the sero-reactor rates increased with age. The oldest persons had infections of much longer duration than the younger individuals when they received treatment at ITS, and serofastness increases with duration of infection when treatment is initiated.

Table XXIV shows the TPI test results (1968) in persons for whom ITS data were available. It is recalled that a subsample of 25 per cent. of all the sera collected during the sero-epidemiological study was tested by TPI. Tests were performed for 690 persons out of the total of 2,402 for whom ITS data were available (28·7 per cent.). The TPI reactivity rate of the group which received case treatment at ITS was 54·4 per cent. (40·4 per cent. reactive, 14·0 per cent. weakly reactive). No person who had received contact treatment was found to be TPI-reactive. In the group which had not received treatment at ITS, only 3·6 per cent. were found to be TPI-reactive (1·5 per cent. reactive and 2·1 per cent. weakly reactive). Furthermore, TPI testing was carried out in 1968 in 1,668 persons for whom no ITS records were then available. Of these 1,668 tests, six were reactive (0·4 per cent.) and seven weakly reactive (0·4 per cent.), the remaining 1,655 all being non-reactive (99·2 per cent.).

It is striking that the majority of the TPI-reactors belonged to the group which had been diagnosed and treated at ITS, although the group-represented only 5 per cent. of the population examined (122 out of 2,402). Persistence of immobilizing antibodies, even 15 to 20 years after adequate treatment, is found in all treponemal infections of some duration. The very few of TPI-reactors found in untreated persons and in the remainder of the population in Bosnia should probably be attributed to infections which had regressed before ITS—either spontaneously or as a result of treatment—and which already had become non-reactive in the Meinicke test. It is also possible

TABLE XXIV *TPI results* (1968) *in* 690 *persons regarding whom ITS records were available, according to decision for treatment*

ITS treatment			*TPI results*					
Decision	*Number*	*Examined in TPI* (1968)	*Reactive*		*Weakly reactive*		*Non-reactive*	
			Number	*Per cent.*	*Number*	*Per cent.*	*Number*	*Per cent.*
Case treatment	122	57	23	40·4	8	14·0	26	45·6
Contact treatment	22	18	0	0	0	0	18	100
No treatment	2,258	615	9	1·5	13	2·1	593	96·4
Total	2,402	690	32	4·6	21	3·0	637	92·3

that latent infections which had remained undetected at ITS because of the low sensitivity of the Meinicke test were brought to light only by TPI testing 15 to 20 years later.

Special clinical and laboratory study of TPI-reactors

In order to define possible neurological, cardiovascular, ophthalmological, or other late complications of endemic syphilis in persons who were found to be TPI-reactive in the evaluation study in 1968, special examinations on such TPI-reactors were carried out in the Institute of Dermato-Venereology at Sarajevo. Of the 66 persons found to be reactive by TPI testing in 1968 in the Sarajevo laboratory, 35 agreed to visit Sarajevo for full examination. All were subjected to a complete cardiovascular, ophthalmological, and neurological examination, including lumbar puncture. Of the 35, twenty had records of adequate treatment with long-acting penicillin at the time of the mass campaign. In these twenty cases, no late complications could reasonably be expected to occur (even if untreated endemic syphilis might have given rise to such complications). As expected, no signs of neurological, cardiovascular, or ophthalmological disturbances which could be ascribed to syphilitic involvement of these organs were found. Of the other fifteen, it was established that eight had not been treated at ITS because no clinical or serological signs of endemic syphilis had been found at that time. Nevertheless, they were found to be TPI-reactive in 1968. For the other seven, no records from the mass campaign could be retrieved, so that it was not known whether they had been treated or not. No unequivocal signs of systemic involvement of the internal organs were found in these fifteen cases. A few showed non-specific neurological or ophthalmological abnormalities, but in the presence of perfectly normal cerebrospinal fluid these could not be accepted as evidence of systemic involvement by treponemal disease.

In order to investigate the presence of spiral organisms, examination of the aqueous humour and cerebrospinal fluid was undertaken in most of this group of 35 TPI-reactors. Some darkfield findings and positive immunofluorescent staining tests showed the presence of such organisms in some of the TPI-reactive individuals, but infectivity tests were negative.

Summary and conclusions

An analysis of data collected in a seroepidemiological study of endemic non-venereal syphilis in Bosnia in 1968 compared with the findings during a previous mass campaign and available post-campaign surveillance data led to the conclusion that transmission of endemic syphilis was completely interrupted by the mass campaign. The latter was carried out against a background of rapid socioeconomic change in the affected population, along with the creation of modern health services to cover the entire population of the Republic of Bosnia-Herzegovina.

Analysis of the background data shows that this excellent result was brought about by intensive treatment of all cases detected through systematic clinical and serological screening of the affected population and of contacts exposed to infectious family members of the initial treatment survey and the re-surveys. Re-surveys during the post-campaign surveillance of the disease covered decreasing numbers of persons and were mainly directed towards high prevalence foci located at preceding surveys. The findings by the TPI test used in the seroepidemiological study in 1968 to 1970 indicate that the diagnostic procedures used during the mass campaign were adequate.

In the introduction to the present report, two essential public health questions were raised in relation to endemic syphilis, notably: '*Can early infectious clinical disease actually be eliminated*' and '*Can transmission of infection be interrupted through the wide application of a specific drug, utilized in accordance with the epidemiological characteristics of the disease?*'

The present study shows that early infectious disease CAN be eliminated and that the transmission of treponemal infection CAN be interrupted by a vigorous and carefully-conducted mass campaign, supported by the organization of an efficient health service with concomitant improvement in socioeconomic conditions, as was the case in Bosnia-Herzegovina.

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Evaluation d'une campagne de masse antérieure contre la syphilis endémique en Bosnie-Herzégovine

SOMMAIRE

Une analyse des informations recueillies dans une étude séro-épidémiologique de la syphilis endémique non vénérienne en Bosnie en 1968, comparée avec les données obtenues lors d'une campagne de masse antérieure et avec les informations disponibles sur le contrôle qui a suivi cette campagne, mène à la conclusion que la dite campagne de masse a complètement interrompu la transmission de la syphilis endémique. Cette dernière s'est développée sur un fond de rapide changement socio-économique de la population affectée, allant de pair avec la création de services de santé modernes intéressant la population entière de la République de Bosnie-Herzégovine.

L'analyse des informations de base montre que ce résultat excellent a dépendu du traitement intensif de tous les cas détectés par un examen clinique et sérologique systématique de la population atteinte et des sujets qui avaient été exposés au contact de parents trouvés infectés lors de l'examen et du ré-examen fait après le traitement initial. Les ré-examens au cours de la surveillance de la maladie après la campagne ont intéressé un nombre décroissant de sujets et ont été particulièrement dirigés contre les foyers de haute prévalence trouvés lors des examens précédents. Les constatations faites à l'aide du test TPI utilisé dans l'étude séro-épidémiologique de 1968 à 1970, indiquent que les dispositifs de diagnostic employés pendant la campagne de masse avaient été appropriés.

Dans l'introduction du présent rapport, deux questions essentielles de santé publique, en relation avec la syphilis endémique, ont été mises en évidence, principalement: "une maladie infectieuse à son stade clinique récent peut-elle être actuellement éliminée?" et "la transmission de l'infection peut-elle être interrompue à l'aide d'une application large d'un médicament spécifique employé d'après les caractéristiques épidémiologiques de la maladie?".

La présente étude montre qu'une maladie à son stade d'infection récente *peut* être éliminée et que la transmission de l'infection tréponémique *peut* être interrompue par une campagne de masse vigoureuse et soigneusement conduite, aidée par l'organisation d'un Service de Santé efficace de même que par une amélioration concomitante des conditions socio-économiques, comme ceci fut le cas en Bosnie-Herzégovine.